REMARKS

The Office Action indicates that Applicants' reply filed on September 22, 2003 is not fully responsive to the prior Office Action because Applicants did not respond to the *Requirement for Information under 37 C.F.R. § 1.105* (see page 6 of the prior Office Action mailed on June 17, 2003). Since the reply to the prior Office Action was otherwise considered *bona fide*, Applicants are given one month or thirty days form the mailing date of this notice to supply the omission or correction. Applicants gratefully acknowledge this invitation and hereby supply the response to the *Requirement for Information*. It is indicated that the omission occurred due to an unintentional error. Entry of the response to the *Requirement for Information* is respectfully requested.

Requirement for Information under 37 C.F.R. § 1.105

The Examiner requests information to account for the two factor difference in SGT specific activity of the instant invention compared to the prior art. Specifically, the Examiner indicates in the prior Office Action that the Applicants have taught purified SGT with specific activities of between 16 to 46 x 10^3 U/mg protein and claimed purified SGT with specific activities of at least about 25 x 10^3 U/mg protein, while the closest prior art found by the Examiner (or provided by Applicants) teaches maximum specific activities that appear to be less than those disclosed by the Applicants by a factor of two. Further, the Examiner points out that Tashiro *et al.* disclose SGT as purified by ion exchange chromatography, just as the SGT fraction in Example 1 of the present invention. According to the Office Action, each of the art references (*i.e.*, Tashiro, Shimura and Kasai, and Kasai) teach similar modes of purifying SGT, or trypsin, but none of them achieve specific purities within two factors of those disclosed by Applicants.

I. Information Provided in Specification

Applicants have addressed the prior art techniques for purifying SGT and their short-comings in the background section of the application (see page 3, paragraph [006] and page 4 paragraph [007]). Therein they state the following (with emphasis added):

"Streptomyces griseus trypsin has been purified by chromatographic techniques using different types of ion exchange resins. These methods typically use stable matrices, which minimize the problem of bleeding of the ligand into the product during elution. **These methods, however, have relatively low selectivity, leading to purification factors in the range of <10. As a result, to achieve a high degree of purity, several steps have to be combined, which in turn may cause autodigestion of the trypsin and therefore loss of activity.** Purification by ion-exchange chromatography on CM-Sephadex, with further purification performed by rechromatography on a ion exchange column has been described by Jurasek *et al.* (1971. Can. J. Biochem. 49:1195-1201) and Olafson *et al.* (1975a. Biochem. 14:1168-1177; 1975b, Biochem. 14: 1161-1167). Miyata *et al.* (1991. Cell Structure and Function 16:39-43) describe a three step cation exchange chromatography process to purify SGT. SGT is found to migrate as a single band in PAGE with a molecular weight of about 30,000 and having an esterase activity higher than bovine trypsin as determined by BAEE assay. **However, even SGT purified by three step chromatography purification methods was found to be slightly contaminated by carboxypeptidase B-like activity.**

SGT has also been purified from Pronase by affinity chromatography using oligopeptides derived from tryptic digest of salmine as highly specific ligand for SGT. Elution of the trypsin-like activity from the mixture of protease in Pronase with HCl revealed purified SGT which was, however, found to be contaminated by carboxypeptidase B-like activity (Kasei *et al.* 1975. J. Biochem. 78.:653-662; Yokosawa *et al.* 1976. J. Biochem. 79:757-763). For analytical purposes only, SGT was also separated from Pronase by affinophoresis using benzamidine as a ligand (Shimura *et al.* 1982. J. Biochem.92:1615-1622)."

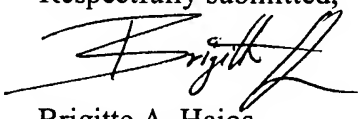
In contrast to the prior art, the present invention provides a *simple large-scale method* for isolation and separation of the trypsin-like fraction of Pronase with high specific activity. Applicants' invention is directed to a method of isolating SGT by a *single chromoatography step* wherein arginine is preferred as an eluant. Applicants employ fewer chromoatography steps then the prior art, thus, the likelihood of contamination is lower, leading to a purer form of SGT and a higher specific activity of the enzyme. Consequently, the yield

obtained by the methods of the invention is higher compared to prior art ion exchange chromatography methods (see page 8, paragraph [029] and page 9, paragraph [030]).

CONCLUSION

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 650-326-2400.

Respectfully submitted,



Brigitte A. Hajos
Reg. No. 50,971

TOWNSEND and TOWNSEND and CREW LLP
Two Embarcadero Center, 8th Floor
San Francisco, California 94111-3834
Tel: 650-326-2400
Fax: 415-576-0300
BAH:klc

60056821 v1

UNITED STATES PATENT AND TRADEMARK OFFICE

Please stamp your date of receipt of the following documents and return to addressee:

First Class Mail Date:	October 31, 2003
Application No.:	10/066,223
Applicant(s):	Mitterer et al.
Attorney:	BAH/klc
Attorney Docket No.:	20695C-002100US

1. Transmittal (1 page)
2. Response to Requirement for Information (4 pages)

60073339 v1